# PATENT COOPERATION TREATY



# PCT

## INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

| Applicant's or agent's file reference | FOR FURTHER ACTION | See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416) |
|---|---|---|
| JF/lh/WCM.103 | | |
| International application No. | International filing date *(day/month/year)* | Priority date *(day/month/year)* |
| PCT/GB 03/04740 | 04.11.2003 | 12.11.2002 |

International Patent Classification (IPC) or both national classification and IPC
C12Q1/00

Applicant
UNIVERSITY OF WALES COLLEGE OF MEDICINE et al.

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 7 sheets, including this cover sheet.

   ☒ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of 3 sheets.

3. This report contains indications relating to the following items:

   - I ☒ Basis of the opinion
   - II ☐ Priority
   - III ☐ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - IV ☒ Lack of unity of invention
   - V ☒ Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   - VI ☐ Certain documents cited
   - VII ☐ Certain defects in the international application
   - VIII ☐ Certain observations on the international application

| Date of submission of the demand | Date of completion of this report |
|---|---|
| 03.12.2003 | 04.02.2005 |

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# INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/GB 03/04740

## I. Basis of the report

1. With regard to the **elements** of the international application *(Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17))*:

**Description, Pages**

1-22 as originally filed

**Claims, Numbers**

1-16 received on 12.01.2005 with letter of 10.01.2005

**Drawings, Sheets**

1/2-2/2 as originally filed

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

   These elements were available or furnished to this Authority in the following language: , which is:

   ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).

   ☐ the language of publication of the international application (under Rule 48.3(b)).

   ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

   ☐ contained in the international application in written form.

   ☐ filed together with the international application in computer readable form.

   ☐ furnished subsequently to this Authority in written form.

   ☐ furnished subsequently to this Authority in computer readable form.

   ☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

   ☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

   ☐ the description, pages:

   ☒ the claims, Nos.: 1-15

   ☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)).

   *(Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)*

6. Additional observations, if necessary:

**IV. Lack of unity of invention**

1. In response to the invitation to restrict or pay additional fees, the applicant has:

   ☐ restricted the claims.

   ☐ paid additional fees.

   ☐ paid additional fees under protest.

   ☐ neither restricted nor paid additional fees.

2. ☒ This Authority found that the requirement of unity of invention is not complied with and chose, according to Rule 68.1, not to invite the applicant to restrict or pay additional fees.

3. This Authority considers that the requirement of unity of invention in accordance with Rules 13.1, 13.2 and 13.3 is

   ☐ complied with.

   ☒ not complied with for the following reasons:

   **see separate sheet**

4. Consequently, the following parts of the international application were the subject of international preliminary examination in establishing this report:

   ☒ all parts.

   ☐ the parts relating to claims Nos. .

**V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement**

1. Statement

| | | | |
|---|---|---|---|
| Novelty (N) | Yes: | Claims | 1, 3-4, 7-8 |
| | No: | Claims | 2, 5-6, 9-16 |
| Inventive step (IS) | Yes: | Claims | - |
| | No: | Claims | 1-16 |
| Industrial applicability (IA) | Yes: | Claims | - |
| | No: | Claims | 1-16 |

2. Citations and explanations

see separate sheet

The following documents are referred to in this communication, the numbering will be adhered to in the rest of the procedure:

D1: GAUCZYNSKI S ET AL: 'Recombinant human prion protein mutants huPrP D178N/M129 (FFI) and huPrP +9OR (fCGD) reveal proteinase K resistance' JOURNAL OF CELL SCIENCE, vol. 115, no. 21, 1 November 2002 (2002-11-01), pages 4025-4036, XP002271831

Section III

1. With respect to claims 1-16 filed with the letter dated 10.01.2005 the attention of the Applicant is drawn to the fact that no unified criteria exist in the PCT for assessment of patentable inventions. The EPO, for example, considers that the whole set of claims, as far as they concern methods of treatment/diagnosis that may be practised on the human or animal body (due to e.g. the step "**obtaining a test sample**" in claim 2), are examined by the IPEA but relate to subject matter considered by the Examining Division at the EPO to be covered by the provision of **Article 34(4)(a)(I) and Rule 67(iv) PCT** and Article 52(4) EPC. Consequently, in an eventual subsequent examination in the regional phase, these inventions would not be considered as being susceptible of industrial application.

Section IV

1. In view of the lack of novelty of newly filed independant claim 2, which claim still relates to a protease digestion with only one protease (see Section V below), present international application is open to objection under Rule 13.1 PCT for the reasons listed below:

2. The only common inventive concept underlying present international application can be seen in the provision of a method to determine the significance of a gene mutation for the protein structure by proteolytic digestion. However, taking into account that such method is already taught in D1 said common inventive concept no longer exists. Correspondingly present claims no longer relate to one invention, thus being in discordance to Rule 13.1 PCT. The opinion of this International examining Authority is that said international application relates to at least two separate groups of inventions, namely:

group I: claims 1-16 (only partially) - a method of determining the structural properties of variants of a protein by exposing it to plurality of proteases;

group II: claims 1-16 (only partially) - a method of determining the structural properties of variants of a protein by exposing it to one protease.

Section V

1. Document D1 discloses a method for the characterisation of the prion protein mutant isoforms (see e.g. Fig.2 and page 4034, right-hand column) by digesting said isoforms with proteinase K, analysing fragment patterns on the western blot in comparison to the wt-PrP. Therefore, present claims 2, 5-6 and 9-16 are not novel over D1 (Article 33(2) PCT).

2. The use of proteases in the characterisation of proteins is, next to the use of restriction endo-nucleases in the characterisation of DNA, one of the initially developed technics in the field of biochemistry and molecular biology. This IPEA considers a method for the determining of a polymorphism or a mutation in a protein, or respectively in its encoding nucleic acid, by using protease(s) to digest said protein, as being obvious for the skilled artisan. Thus, present claims 1, 3-4 and 7-8 cannot be acknowledged for involving an inventive step (Article 33(3) PCT).

3. The only claims that may be considered as being novel are present claims 1, 3-4 and 7-8, which relate to the use of more than one protease. Yet, it remains obscure how to perform the subject matter of said claims for instance in the case where said proteases are used simultaneously. The present Application do not provide with a **technical** support in the description with respect to the simultaneous treatment with proteases. Hence, the subject matter of these claims can be seen only as being a scientific theory, thus having no industrial applicability.

4. For the sake of completeness it is noted that expressions like "conventional protein assay" and "additional studies" as in present claims 11 and 14, are so broad that it is not possible to clearly determine the subject matter for which protection is sought (Art. 6 PCT).

5. It is also noted that in view of present claim 1 claim 3 seems to be redundant (Article 6 PCT).

CLAIMS

1. A method for determining the significance of a given nucleic acid polymorphism or mutation, in a nucleic acid molecule, on the structural properties of a protein encoded by said nucleic acid molecule comprising:

(a) exposing the protein encoded by said nucleic acid molecule to a plurality of proteases; and

(b) determining whether, or to what extent, proteolytic cleavage takes place; and, optionally,

(c) comparing this proteolytic cleavage with that of the wild-type protein when exposed to the same protease(s).

2. A screening method for determining the significance of a plurality of variants of at least one gene comprising:

(a) obtaining a sample of protein encoded by each of said variants;

(b) exposing each protein to at least one protease;

(c) determining whether, or to what extent, proteolytic cleavage takes place; and

(d) comparing the proteolytic cleavage with that of the wild-type protein when exposed to the same protease(s).

3. A method according to claim 2 wherein said protein is exposed to a plurality of proteases.

4. A method according to claim 3 wherein at least some of said proteases attack different sites within the protein.

5. A method according to any preceding claim wherein said protease(s) comprises any one or more of the following: trypsin, chymotrypsin, proteinase K, aminopeptidase, carboxypeptidase, collagenase, elastase, Kallikrein, metalloendopeptidase, papain or pepsin.

6. A method according to any preceding claim wherein a plurality of proteins are exposed to said protease(s).

7. A method according to claims 3-6 wherein said proteins are exposed to said proteases, or vice versa, simultaneously.

8. A method according to any preceding claim wherein said protein(s) is exposed to said different proteases either simultaneously or successively.

9. A method according to any preceding claim wherein said protein(s) are exposed to said protease(s) under conditions that support the activity of said protease(s).

10. A method according to any preceding claim wherein digestion of said protein(s) is terminated by adding at least one protease inhibitor to the reaction.

11. A method according to any preceding claim wherein proteolytic cleavage is determined using a conventional protein assay.

12. A method according to claim 11 wherein said assay involves SDS-PAGE analysis.

13. A method according to claim 12 wherein said analysis is followed by staining or blotting.

14. A method according to any preceding claim wherein additional studies are undertaken to determine the functionality of the protein variant.

15. A method according to any preceding claim wherein part (a) involves further exposing the wild-type protein to said at least one protease and part (b) involves determining whether and to what extent proteolytic cleavage of said wild-type protein takes place.

16. A method according to any preceding claim wherein the wild-type protein and, optionally, the variant protein are subjected to the conditions of the proteolytic reaction, in the absence of the said protease(s), and then the extent of proteolytic cleavage is determined.